**Electronic Supplementary Material**

This is the ESM to the article

**ENVIRONMENTAL RISK FACTORS ASSOCIATED WITH BOVINE TUBERCULOSIS IN CATTLE IN HIGH RISK AREAS**

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**Descriptive statistics from logistic regression analysis of the TB99 study and Akaike’s information statistics when the TB99 model was applied to the CCS05 study**

**Descriptive statistics from logistic regression analysis of the TB99 study**

The Tables 1 and 2 contain the descriptive statistics for the variables considered to be included in the model when analysing the TB99 dataset. All variables that had a significance (*P* <0.10) were considered for inclusion in the model, except those where high numbers of missing values made the models unstable and prevented multi-model comparisons. These variables with high missing values included the perceived presence of badgers on the farm and spreading of manure and slurry.

We did not include parish testing interval in the TB99 model because the case and respective control herds were selected from the same area and most herds in the TB99 study were situated in areas with 1 year testing interval. We did test the variable and it did not significantly affect the model. We opted to use a single variable to describe animal movement (movement of cattle on) and herd contacts (contiguous contacts). Strip grazing was omitted from the final model because it had a positive interaction with herd type, being used mainly on dairy farms. Similarly, cattle epidemiological group, stocking density and grazing area interacted with herd size category. The Pearson’s correlations between the variables included in the models ranged from r = -0.01 to 0.12. Mineral supplementation was tested to see if supplemented herds had a lower risk of acquiring bTB. However, very few farms supplemented their herds with minerals and a more accurate recording is needed to test effectively mineral status of the herds. We decided not to include housing types and characteristics, such as the height of troughs, in the analysis. Clay and loam soil were the only soil characteristics that had enough variation to be included in the model. Clay soil was not significant in univariate analysis and loam soil lost significance when included in the main model. There was no interaction between the environmental variables in the model. There was no interaction between maize, silage and grazing silage-hay aftermath and between those variables and herd size and type. Area planted with maize interacted negatively with area of deciduous woodland. When the interaction was included in the main model it lost its significance, so no interactions were included in the main model.

The effect of the herd previous bTB status was tested on a subset of control herds that had not suffered a previous breakdown and their respective case herds (see Table 3).

**Table 1. Number of case and control farms for the binary variables tested in the study (all farms), odds ratio and significance (*P*) when the variable was included in the top ranking modela**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Variable | Case Farms (n) | | Control Farms (n) | | Odds ratio | *P* |
| negative | positive | negative | positive |
|  |  |  |  |  |  |  |
| Closed herd | 334 | 169 | 535 | 273 | 0.80 | 0.11 |
| Organic certification | 466 | 37 | 741 | 67 | 1.25 | 0.35 |
| Strip grazing | 245 | 258 | 399 | 409 | 0.76 | 0.06 |
| Set grazing | 340 | 163 | 558 | 250 | 0.88 | 0.32 |
| Sown pastures | 256 | 247 | 456 | 352 | 1.01 | 0.96 |
| silage feeding | 38 | 465 | 158 | 650 | 2.18 | 0.00 |
| Grazing silage/ hay aftermath | 193 | 310 | 247 | 561 | 0.56 | 0.00 |
| Grazing woodland | 472 | 31 | 777 | 31 | 1.37 | 0.26 |
| Grazing scrub | 495 | 8 | 797 | 11 | 0.89 | 0.83 |
| Grazing moorland pasture | 498 | 5 | 799 | 9 | 0.51 | 0.27 |
| Supplement General Mineral | 496 | 7 | 800 | 8 | 1.26 | 0.67 |
| Supplement Dry Cow Mineral | 502 | 1 | 807 | 1 | 0.86 | 0.92 |
| Selenium supplementation | 484 | 19 | 776 | 32 | 0.88 | 0.70 |
|  |  |  |  |  |  |  |

aThe final model contained the variables incident number, cull areas, herd size category, enterprise type, grazing area, number of premises, maize (ha+), deciduous forest (ha), marsh area (ha), rough pasture (ha), movement of cattle on to farm, internal boundaries hedge (%), grazing silage hay aftermath, mean number of cattle in epidemiological groups, silage feeding

**Table 2. Number of case and control farms of the multi-level categorical and numerical variables, odds ratio and significance (*P*) when the variable was included in the final modela**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Variable | Case Farms (n) | Control Farms (n) | Odds ratiob | *P* |
| **Categorical variables** |  |  |  |  |
| Main type of Enterprise |  |  |  |  |
| Beef | 191 | 355 | - | - |
| Dairy | 244 | 247 | 1.23 | 0.20 |
| Sheep | 32 | 67 | 1.12 | 0.66 |
| Other | 36 | 139 | 0.47 | 0.001 |
| Herd size category |  |  |  |  |
| Large (>150 cattle) | 238 | 268 | - | - |
| Medium (50 to 150 cattle) | 192 | 293 | 0.98 | 0.89 |
| Small (<50 cattle) | 73 | 247 | 0.57 | 0.016 |
| Cull areas |  |  |  |  |
| Control | 169 | 275 | - | - |
| Pro-active | 176 | 267 | 1.08 | 0.60 |
| Reactive | 158 | 266 | 0.71 | 0.82 |
|  |  |  |  |  |
| **Numeric variables** |  |  |  |  |
| Number of premises | 2.02 | 1.77 | 1.06 | 0.24 |
| Cattle epidemiological groups (n) | 43.18 | 31.69 | 1.01 | 0.014 |
| Stocking density (n/ha) | 6.51 | 4.42 | 1.01 | 0.21 |
| Movement of cattle on (n) | 21.32 | 41.10 | 1.00 | 0.00 |
| Contiguous contacts (n) | 9.63 | 8.38 | 1.00 | 0.575 |
| Grazing area (ha) | 43.98 | 37.05 | 1.00 | 0.016 |
| Deciduous woodland (ha) | 2.94 | 1.55 | 1.04 | 0.002 |
| Coniferous woodland (ha) | 0.12 | 0.15 | 1.00 | 0.906 |
| Marsh area (ha) | 1.04 | 0.36 | 1.06 | 0.012 |
| Rough pasture (ha) | 5.99 | 2.51 | 1.01 | 0.023 |
| Internal boundaries – hedge (%) | 0.67 | 0.74 | 0.64 | 0.002 |
| Maize area (ha) | 3.72 | 1.85 | 1.02 | 0.077 |
| Cereal area (ha) | 14.25 | 13.24 | 1.00 | 0.558 |
| Set aside (ha) | 0.69 | 0.62 | 1.00 | 0.830 |
| Clay soil (ha) | 28.48 | 25.52 | 1.00 | 0.130 |
| Loam soil (ha) | 49.75 | 38.77 | 1.00 | 0.207 |
|  |  |  |  |  |

aThe final model contained the variables incident number, cull areas, herd size category, enterprise type, grazing area, number of premises, maize (ha+), deciduous forest (ha), marsh area (ha), rough pasture (ha), movement of cattle on to farm, internal boundaries hedge (%), grazing silage hay aftermath, mean number of cattle in epidemiological groups, silage feeding

bLevels with no odds ratio and significance values, were used as the reference level.

**Analysis of TB99 dataset only including control farms that had no previous breakdown**

Considering the period of 12 months without breakdown for the selection of controls herds and therefore trying to account for any possible residual effect of breakdown before that period, we have repeated the analysis of the TB99 dataset only including control herds that did not have a previous breakdown. The results are very similar to the results when the whole dataset is analysed. The odds are slightly smaller corresponding to the smaller number of farms.

**Table 3. Predictor weights and model averaged odds ratios of variables appearing in the top models from logistic regression of bovine tuberculosis incidence – farms with no previous breakdowns**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Variable** | **Number of models in which variable appears**  **(out of 4)** | **Predictor weight** | **Odds ratio from multivariate model** | **95% CI for multivariate odds ratio** |
| Deciduous wood (10 ha) | 8 | 0.94 | 1.37 | 1.03 – 1.90 |
| Marsh (10 ha) | 7 | 0.83 | 1.67 | 0.90 – 3.80 |
| Rough pasture (10 ha) | 7 | 0.82 | 1.07 | 0.99 – 1.20 |
| Internal boundary hedge (%) | 8 | 0.94 | 0.68 | 0.47 – 0.94 |
| Maize (10 ha) | 6 | 0.76 | 1.20 | 0.98 – 1.65 |
| Grazing silage hay aftermath (yes/no) | 9 | 1.00 | 0.53 | 0.38 – 0.73 |
| Feeding silage (yes/no) | 9 | 1.00 | 2.64 | 1.60 – 4.35 |
| Herd size category:  Small (<50 cattle)  Medium (50-150 cattle)  Large (>150 cattle)1 | 9 | 1.00 | 0.35  0.65  - | 0.21 – 0.59  0.44 – 0.97  - |
| Cattle enterprise type:  Beef1  Dairy  Sheep  Other | 9 | 1.00 | -  1.06  1.13  0.41 | -  0.71 – 1.58  0.62– 2.06  0.24 – 0.72 |
| Cattle moving on (10 cattle) | 9 | 1.00 | 0.94 | 0.91 – 0.97 |
| Cull areas:  control1  reactive  pro-active | 1 | 0.06 | -  1.02  1.07 | -  0.72 – 1.52  0.77- 1.66 |
|  |  |  |  |  |

**Akaike’s statistic when the model obtained from the analysis of the TB99 study was applied to the CCS05 study**

The CCS05 case-control study was set up in areas with moderate to high incidence levels of bTB and in areas of the country that differed in management and environmental farm characteristics when compared to the RBCT areas. Furthermore, the selection of controls was more rigorous in the CCS05 study and the questionnaire and data gathering was altered since the TB99 study (6). Any of those factors could have influenced the comparison of results between the 2 studies. The main differences in the results from the two datasets were: 1) dairy farms increased the odds for breakdown in the analysis of the TB99 study and in the analysis of the CCS05 study it decreased the odds for breakdown; 2) the effect of feeding silage was much smaller in the CCS05; 3) seasonally wet soils did not alter the odds ratio of bTB in the CCS05 study, but increased the odds for breakdown in the TB99 study - however different kinds of assessment were used for this variable (marsh areas and seasonally wet soils could potentially represent areas with different degrees of wetness, marsh areas being wet all year round and seasonally wet soils only part of the year) ; 4) area of woodland decreased the odds ratio for bTB breakdown and increased in the TB99 study; 5) percentage of boundaries comprised of hedges did not alter the odds ratio for BTB breakdown in the CCS05 dataset. The results on woodland and hedges probably reflect the different role that badgers play in the disease transmission in areas with different prevalence of the bTB.

**Table 4. Akaike information statistic ranking logistic regression mixed models containing herd and habitat variables that affect the incidence of bovine tuberculosis on cattle farms CCS05 trial (10 higher ranking models out of 35)**

|  |  |  |  |
| --- | --- | --- | --- |
| **Model** | **AIC** | **∆AIC** | **Akaike weight** |
| Woodland area ha, Seasonally wet soil ha, Rough pasture ha, Maize (y/n), Feeding silage (y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1477.02 | 0.00 | 0.08 |
| Woodland area ha, Rough pasture ha, Maize (y/n), Feeding silage (y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1377.03 | 0.01 | 0.08 |
| Woodland area ha, Rough pasture ha, Maize ha(y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1477.23 | 0.21 | 0.07 |
| Woodland area ha, Seasonally wet soil ha, Rough pasture ha, Maize (y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1477.31 | 0.29 | 0.07 |
| Woodland area ha, Seasonally wet soil ha, Rough pasture ha, Maize (y/n), Feeding silage (y/n), N. cattle, Herd type, Incident number | 1477.75 | 0.73 | 0.06 |
| Woodland area ha, Rough pasture ha, Maize (y/n), Feeding silage (y/n), N. cattle, Herd type, Incident number | 1477.88 | 0.86 | 0.05 |
| Woodland area ha, Seasonally wet soil ha, Rough pasture ha, Maize (y/n), N. cattle, Herd type, Incident number | 1478.03 | 1.01 | 0.05 |
| Woodland area ha, Rough pasture ha, Maize (y/n), N. cattle, Herd type, Incident number | 1478.07 | 1.06 | 0.05 |
| Woodland area ha, Seasonally wet soil ha, Rough pasture ha, Maize (y/n), Hedges (%), Feeding silage (y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1478.90 | 1.89 | 0.03 |
| Woodland area ha, Rough pasture ha, Maize (y/n), Hedges (%), Feeding silage (y/n), N. cattle, N. cattle added to herd, Herd type, Incident number | 1478.92 | 1.90 | 0.03 |

AICc – Akaike information criterion, ∆ AICc – indicates the amount of support for the model relative to the top ranking model, Akaike weight - The Akaike weights can be interpreted as the probability of the candidate model being the ‘best’ out of all those considered.

**Table 5. Predictor weights for variables appearing in the top models and odds ratios from logistic regression of bovine tuberculosis incidence – CCS05 trial**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Variable** | **Number of models in which variable appears (out of 35)** | **Predictor weight** | **Univariate odds ratio** | **Odds ratio from multivariate model** | **95% CI for odds ratio** |
| Woodland area (10 ha) | 32 | 0.96 | 0.98 | 0.97 | 0.95 – 1.00 |
| Seasonally wet soils (10 ha) | 17 | 0.50 | 1.00 | 1.00 | 1.00 – 1.01 |
| Rough pasture (10 ha) | 27 | 0.88 | 1.10 | 1.09 | 1.01 – 1.19 |
| Hedge (%) | 8 | 0.20 | 1.00 | 1.00 | 0.99 – 1.01 |
| Maize(yes/ no) | 27 | 0.88 | 0.82 | 1.46 | 1.03 – 2.06 |
| Feeding silage (yes/ no) | 18 | 0.53 | 0.39 | 0.58 | 0.28 – 1.23 |
| Cattle kept at farm (10 cattle) | 35 | 1.00 | 0.98 | 0.98 | 0.96 – 0.99 |
| Cattle enterprise:  Beef  Dairy  Sheep  Other | 35 | 1.00 | -  0.48  1.71  1.34 | -  0.57  1.71  1.27 | -  0.42 – 0.77  0.97 – 3.01  0.66 – 2.47 |
| Cattle added (10 cattle) | 19 | 0.61 | 0.98 | 1.02 | 1.00 – 1.04 |
|  |  |  |  |  |  |

Cattle enterprise – odds ratio compared to beef farms